VOLUME LIV NUMBER 3

THE
BOTANICAL GAZETTE

SEPTEMBER 1912

THE LIFE HISTORY OF ANEURA PINGUIS

CONTRIBUTIONS FROM THE HULL BOTANICAL LABORATORY 159

GRACE L. CLAPP

(WITH PLATES IX–XII)

Historical

In his classical study of liverworts, LEITGEB (**19**) has given a comparative treatment of the genus *Aneura*. Stages of development chosen from several species picture the life history of the genus rather than that of any one species from spore to spore. HOFMEISTER (**13**) earlier described the apical cell and sex organs of *Aneura pinguis*, and KNY (**15**) worked out in an elaborate scheme the segmentation of its apical cell. LE CLERC DU SABLON (**18**), GOEBEL (**10–12**), CAMPBELL (**3**), and CAVERS (**4**) have since added facts, but gaps have been left in the continuous development, chiefly in the embryogeny and in the growth of the sporeling.

Material

Material of *Aneura pinguis* was collected by Dr. LAND at Xalapa, Mexico, in the autumns of 1906, 1908, and 1910. The region around Chicago has offered abundant supply for field study. The plants were killed in the field in Flemming's fluid (weaker), in alcohol (50 per cent) and formalin, and in chrom-acetic acid without osmic acid. Following the close series of alcohols in dehydration, the material imbedded in paraffin was cut in sections 3–10 μ thick. Safranin and anilin blue, Haidenhain's iron-hematoxylin with and without Magdala red, and Flemming's triple stain were used for

stains. Shrinkage in the young embryo and resistance to infiltration of the mature capsule were the chief difficulties met.

Thallus

SCHIFFNER (**23**) describes *Aneura pinguis* as a cosmopolitan species, strictly dioecious. LAND reports the growth of the species on decayed fallen logs in the rain forest as most luxuriant. The plants exceed only slightly, however, those growing in the hydromesophytic habitats of Chicago. On fallen hemlock logs in shady ravines and on the mossy edges of pine-oak dunes bordering sloughs the plants form a close mat. Wherever moss forms a humus layer along the lines of seepage of the clay bluffs along Lake Michigan, the thalli are also found. In the prairie meadows *Aneura pinguis* often extends half an inch on *Typha* and *Acorus* leaves or grows on clumps of exposed grass roots. In all these places moisture, diffused light, and fairly low temperature are the favorable conditions for its growth. Plants grow fairly well in the laboratory when these conditions are imitated.

Aneura pinguis is strictly dioecious, but both kinds of plants grow side by side in the field, often with margins overlapping (fig. 1). In general the thallus is a flat ribbon-shaped plant closely appressed to the ground, with frequent branches, and slight indentations along the margins. While it averages 5–7 mm. in width, it may be reduced when growing in a moist chamber with light from one side in the laboratory to less than 2 mm. (NĚMEC **21**).

The thallus consists of compact cells, unequally elongated in the direction of the main axis of the plant. Although it lacks a midrib, the plant is usually thicker along the center, thinning out toward the lateral margins. All the cells contain chloroplasts at some time and the plastids often have 5 or 6 starch grains. Ordinarily it is 10–12 cells thick. There is no definite differentiation into tissues, but the superficial layer is clearly composed of smaller cells with a larger number of chloroplasts. This dorsal "small-celled epidermis" has undergone one more division, longitudinally and transversely vertical, than the layers beneath (fig. 2). Plants in the shade appear succulent and are deep emerald green in color. The numerous chloroplasts are found dividing as often as in *Funaria* or *Elodea*.

On the ventral surface rhizoids (figs. 13–15) and mucilage hairs (fig. 12) indicate slight cell differentiation. The rhizoids contain chloroplasts at first like the other superficial cells (fig. 13). Such cells project slightly, elongate, the chloroplasts disappear, and the rhizoids look like root-hairs. As they grow longer the position of the nucleus changes and the cytoplasm varies in amount and distribution. Both are near the somewhat thickened tip when the rhizoid is old. The rhizoids resemble root-hairs in that they become irregularly lobed in contact with soil particles (fig. 15) and flatten out in a most deformed way against other thalli and bark. BOLLETER (**2**) notes this same lobing in *Fegatella conica* and it is common among liverworts. The length and number of rhizoids varies greatly. In contact with an underlying thallus or bark they are very short (0.09–0.16 mm.); in soil and moist air they average less than 1 mm., and only occasionally reach 2 mm. On closely appressed plants the rhizoids are numerous and scattered; in soil they grow along the central axis but are never definitely localized. Irregularity may come from fungi in the rhizoids, and again they may be as straight as if uninfected. In no case were the lobes of the rhizoids cut off by walls.

Gemmae have been described by many for *Aneura pinguis*. In the material at hand no gemmae were found. If they are characteristic of this species, their absence must be due to the conditions under which the plants grew. EVANS (**7**) has found in species of *Metzgeria* that gemmae are not likely to appear when the plant is growing luxuriantly. This would account for their absence in the field, but one might expect them to appear on plants grown under less favorable conditions in the laboratory.

Increase in the number of plants is brought about, as in many other thallose liverworts, by the dying away of older parts, when branches become the main axes of new individuals.

Aneura pinguis produces three kinds of branches, the ordinary vegetative ones and those bearing the two different sex organs. Such branches have their origin in the segments of the apical cell. All of the descriptions of this cell agree that it cuts off segments on two sides alternately right and left (figs. 3–11). Vertical sections through any of the marginal indentations which indicate

growing points show clearly that its longer axis is the vertical one (fig. 2).

Very rarely at the forward end of the thallus can only one apical cell be found (figs. 5, 6). Usually two indentations are separated by a narrow marginal projection and each sinus contains two apical cells (figs. 9–11). One set carries on the main axial growth, the other produces a branch. The apical cell of the branch originates in a segment of the axial apical cell by a curved vertical wall bent to the right or left so that it strikes the wall of the segment opposite the last cutting one.

The more rapidly growing dorsal surface carries the apical cell to the ventral side (fig. 2). The wall-formation in either direction, therefore, shows an obliquity which later is distinctly vertical or horizontal. The primary segment is divided by a vertical transverse wall into an inner posterior cell and an outer anterior marginal one. Walls parallel to the surface come in now, followed by more vertical ones. The order of vertical, transverse, or longitudinal does not seem fixed. Clearly these three directions give thickness, length, and width to the thallus.

If the growth of two apical regions is nearly equal, the thallus appears dichotomously branched; if one grows faster, the other is recognizable as an indentation on the lateral margin, where it may remain growing very slightly if at all; or if the main axis be injured, it becomes the apical region of the main thallus, growing rapidly.

Characteristic of the apical cell are the mucilage hairs borne on the ventral surface of the thallus (fig. 12); 6–10 of them curve inward and upward around the growing point. KNY and LEITGEB describe the mucilage cells as bearing no direct relation to the thallus in arrangement. They show an alternation, however, corresponding to that of the apical cell segmentation. The superficial cell from which the hair originates first projects from the surface, and then divides into two cells. The basal inner cell retains its plastids permanently; the chloroplasts of the outer one, after some growth, are transformed into a mucilaginous stuff, which stains very deeply. These are the hairs which are sloughed off as the thallus grows. The basal cell divides like any superficial cell. Apparently it is the posterior ventral surface cell, cut from the

primary segment by a vertical transverse and horizontal wall, which produces it.

Sex organs

ANTHERIDIA.—The antheridia of *Aneura pinguis* are borne in the upper surface of lateral branches which occur singly, in groups of three, or occasionally in groups of two. On the branch their arrangement is extremely regular, in two alternating rows corresponding to the segments of the apical cell. They appear imbedded because of the rapid marginal growth of the surrounding cells.

LEITGEB'S diagram for their arrangement in *Aneura palmata* holds true also for *Aneura pinguis*. After the division of the primary segment into an outer marginal and an inner posterior cell by a vertical transverse wall, and after horizontal cleavage of the latter into a dorsal and a ventral cell, the dorsal cell by a vertical longitudinal division forms an inner (toward the median axis of the thallus) and an outer (toward the lateral margin) cell. The inner by a transverse vertical cut divides into two cells, the anterior of which gives rise to the antheridium. This dorsal superficial cell (the antheridium initial), containing a large nucleus and abundant cytoplasm, enlarges and projects (fig. 16). It divides into two cells, the outer of which, by a horizontal wall, forms a stalk cell usually dividing once at least, and the antheridium mother cell (fig. 17).

The method of development follows the *Jungermannia* type. First a vertical wall divides the outer cell into two equal halves (fig. 18). By two vertical intersecting walls in each half, a wall layer of four cells surrounds two central cells—primary spermatogenous cells (figs. 22, 23). By rapid growth the antheridium becomes spherical and appears transparent (fig. 26). Its wall cells, however, contain chloroplasts which persist until the sperm mother cells are distinguishable. The wall cells of the upper half grow noticeably larger than those of the lower half.

No definite cytological study of spermatogenesis was made, but some few points were noted. No centrosome was evident during any division of the spermatogenous cells. The diagonal division of the sperm mother cells separates them by a membrane which

stains as deeply as does the cell wall of the mother cell. The oval nucleus stains deeply and soon occupies one end of the protoplast (fig. 27). At the other end, a dark spot appears in the cytoplasm—the blepharoplast—which gives rise to the cilia. The nucleus in its growth elongates, soon making a turn around the cytoplasm. The cilia are not easily distinguished from the coils of the nucleus. Beside the beak of cytoplasm, anterior to the nucleus from which the cilia extend, a rounded mass is left at the posterior end of the sperm. This is a mechanical hindrance to the movement of the sperms when the cell walls are transformed into a mucilaginous substance and the sperm is often twisted into small spirals on its own axis (fig. 27). These disappear as soon as space is given, and at the time of shedding the mass of cytoplasm at the base is also gone. The body of the sperm is very long and averages about 2.5 times that of the cilia. Cilia 35.2 μ and sperm 88.7 μ, in rough figures, were the measurements of the longest sperms squeezed out of the antheridium before it had burst naturally. Probably when shed in the field they have grown somewhat longer.

The antheridia begin to form early in the spring. They develop in acropetal succession until August, when many of the branches, as has been noted (LEITGEB, CAMPBELL), continue vegetative growth. The last antheridium formed is sometimes not imbedded, but superficial, owing to the rapid elongation of the thallus.

ARCHEGONIA.—*Aneura pinguis* bears its archegonia also on the dorsal surface of distinct lateral branches (fig. 28). Such plants have conspicuously light green filamentous outgrowths, varying in length and width on the lateral margins. These are caused by the more rapid growth of the thallus edges of the main axis or of the lateral branch. Again, there may be from one to three branch primordia; usually, however, one outstrips the others in development. Like the antheridia, the archegonia are regularly arranged in two rows, alternating according to the apical cell segmentation. The division of the primary segment is as usual. The first dorsal superficial cell is the archegonium initial. In order of development it follows the general liverwort type. Three vertical intersecting walls surround a central cell from which a cap cell is cut off by a horizontal wall (fig. 29). The central cell gives rise to four neck

canal cells, a ventral canal cell, and the egg cell. The archegonium wall has two layers of cells (figs. 32, 33). The 4–6 canal cells are of short endurance; their walls break down and the cytoplasm and nucleus are transformed into a mucilaginous substance. The egg cell is large and round, its cytoplasm containing many starch grains (fig. 33). When the cap cells bend back, there is a clear passage made in the neck to the egg. Fertilization was not observed.

As soon as fertilization has occurred, the neck and venter cells divide rapidly, and the whole branch is a thick cushion of cells projecting beyond the margin of the thallus. Usually only one embryo grows on a branch, and where two appear they probably belong to two branches. Two have been reported, however, in one calyptra (COKER **6**). Occasionally archegonia, immature and after the egg has been fertilized, are carried with the filaments to the top of the calyptra in its growth. LEITGEB thinks more trichomes are produced on the torus, but this seems unlikely, for many are sloughed off as it develops. Some new rhizoids do grow at the bulbous base.

Sporophyte

The first division of the egg is a transverse one into epibasal and hypobasal cells (fig. 34). The hypobasal cell has been said either to form a few divisions or to grow into a lobed haustorial cell (LEITGEB **19**). It distinctly becomes a true haustorium (figs. 35, 36), rhizoidal in form. Both cells elongate rapidly; the haustorium sometimes lobes and sometimes remains straight. The epibasal cell is divided by a horizontal wall into two cells (fig. 35) containing abundant cytoplasm, many plastids, and large nuclei. In this three-celled stage disorganization of the cells of the calyptra around the base of the suspensor is striking. The uppermost cell divides again by a horizontal wall, so that a filament of four cells is formed, including the haustorium. Vertical walls now come in, so that there are three rows of quadrants (fig. 36).

The lowest tier next the haustorium now forms two rows (figs. 37, 38), its vertical and transverse walls having no definite sequence. It corresponds to the foot, and the cells form at first a more compact

mass than the slender ones above. The uppermost cell arising in the epibasal row divides to form the capsule; the middle originates the seta, probably by its intercalary growth as LEITGEB has suggested. When, therefore, the seta consists of three or four tiers of cells, the capsule is definitely differentiated. It consists of two rows, each of eight cells. Periclinal walls have cut out a wall layer one cell thick (fig. 38), leaving a central sporogenous tissue of eight cells. The lower four divide by horizontal and vertical walls; the upper also divide, but form only a group of sterile cells—a cap, later continuous with the elaterophore. The wall of the lower half becomes two layers by periclinal divisions not at all simultaneous (figs. 39, 40). The difference in rate of growth from now on is a striking feature of development in the capsule (LE CLERC DU SABLON 18). It first shows in the contrast between the lower and peripheral region and the upper central part. The contents of the cells differ in size of nuclei and amount of cytoplasm. Cell divisions are in every direction. The capsule changes from spherical to oval and elongates rapidly (fig. 41). This difference in rate of growth accompanies the formation of the elaterophore, but what determines the rate? The more slowly dividing cells of the upper central region begin to elongate, and the elaterophore is outlined (fig. 42); its cells have smaller nuclei and less cytoplasm. Although cell divisions are fewer along its margin, they must still be considered sporogenous tissue. Diagonal divisions and radial arrangement of diamond-shaped cells indicate elongation of the capsule. Cells continuing the axis of the capsule between the elaterophore and the base are still rectangular, like those of the elaterophore except in size of nuclei.

Differentiation among the spindle-shaped cells is the next evidence of separation of the sporogenous tissue into elaters and spore mother cells (figs. 43–50). While in the central axis the elongated diamond-shaped cells appear to form continuous rows to the base, in the radial peripheral regions this is more uncertain. The next stage, and a most unsatisfactory one for study, shows a partial transformation of the walls of the elaters and of the spore mother cells. The elaterophore forms a central cylinder of long prosenchymatous cells, the marginal ones of which have a free tip.

They contain plastids with starch. The protoplasts of the elaters and spore mother cells are outlined by a definite membrane at a distance from the wall. The space between, however, shows less well defined strands. The nucleus of the elater is large (figs. 44, 45), at the center, extending well across the diameter of the cell. There are plastids in the elaters, and BOLLETER (**2**) considers the elaters in *Fegatella* feeders of the spore mother cells because the starch disappears from the elaterophore about this time. The spore mother cells also have very large nuclei and the form of the cell is irregularly rectangular to triangular.

The difference in rate of growth noted before between the peripheral and central regions is much more evident at this stage. The four lobes of the spore mother cells are well rounded toward the outer portions of the capsule, while those at the center are just beginning to be distinct. The nucleus with a clear nucleolus lies at the center of the lobed cell (fig. 46). Two successive divisions of the nucleus form the tetrad of spores.

The cell plate becomes continuous with the deeply staining membrane of the lobes. This membrane soon differentiates into another substance, being added to from the interior where its outline is very irregular. Centrally between the two margins staining indicates lines of some substance which grow out to the outer margin, forming at first irregular projections. Meanwhile, within the protoplast a cellulose layer forms. When the spore is mature the two wall layers are not distinct (figs. 51, 52). The protoplast containing chloroplasts seems to be surrounded by a single brown wall with echinate projections. During this time the elaters have changed. The cytoplasm has come to form a spiral along the wall and a broad brown thickening takes its place (fig. 52). Two spirals are not rare and the elaters are often branched (JACK **14**). Probably examination of the chemical changes taking place in the spore coat would find them similar to those BEER (**1**) has found for *Riccia*.

The cell walls of the elaterophore are thickened in a narrow single spiral. The two wall layers have ring thickenings in the sterile cushion at the apex, and in the two upper layers of cells of the seta irregular thickenings are found. The lines of dehiscence are

remarkably distinct in cross-section (fig. 54), for the walls do not change on that side, but remain thin, composed of cellulose.

The seta, measuring about 2 mm. in length, would be described as having a club-shaped foot if it can be called a foot. Even when the seta consists of a few tiers of cells, the glandular appearance at the base is striking. Tissues of the calyptra and seta disorganize so that at the base of the seta there are always some glandular cells and others very much crushed. During its growth the bulbous base of the gametophyte and sporogonium has turned from a horizontal to a vertical position.

The capsules dehisce progressively along the thallus from early spring (March) through May. GOEBEL (**10**) has well described the dehiscence and shedding of spores in *Aneura palmata*. The seta elongates rapidly (NĚMEC **20**) from 2 to 30 and more mm., in the field often twisting on its own axis. Individually its rectangular cells lengthen from 60 μ to 500 and 600 μ. This pushes the capsule far beyond gametophyte and calyptra. Along the well marked lines between the valves, about one-third of the way from the tip at the greatest width of the capsule, a splitting begins. The crack lengthens until with a jerk the valves are bent back. Some spores are freed now, but the majority are shed by the next movement of the valve, when its fourth of the elaterophore springs upward 45° or more. Spores and elaters fall together, the tetrad often complete.

Germination of spores

Plants with capsules about to shed were brought from the field March 23, April 15, May 17, and May 20, and put on wet cotton under bell-jars or in large Petri dishes. Spores were sown as soon as the capsules burst. On sterilized cotton the spores (averaging 60–68 or 70 μ) are soon lost. A better medium and more easily examined under the microscope is made by putting a layer of heavy white filter paper over wet cotton in a Petri dish. Porous clay plates are also good. Drop cultures in 0.5 and 1 per cent cane sugar, 2.5 and 3 per cent glucose, 0.5 and 1 per cent lactic acid, 0.3 and 0.6 per cent Knop solution, vegetable lipase, distilled water, all died after reaching the two-celled stage. The excessive amount of moisture was one cause of this, for cultures made at the

same time on cotton with distilled water and 1 per cent cane sugar lived. On moist cotton in the sunlight the spores died in the one-celled or two-celled stage. Other cultures, therefore, were kept in a room with the window open, so that the temperature varied roughly with that outside, and the light was kept diffuse by the window shade. Sowings were made on sterilized clay, sand, sphagnum, humus, and sand, and kept under bell-jars. Cultures on rotten wood were spoiled by *Pencillium*. Although the pots containing the soils were scrubbed, dried, sterilized over night in a drying oven above 115° and again with the soils in an autoclave, many became infected with a species of *Chaetomium*. This could have come about when spores were taken out for examination. The accompanying table records some of the data.

Sowing	Final examination	Time	Medium	Condition of spores
March 23...	June 23	3 mos.	H_2O on cotton	2-celled to all stages (fungi)
April 15...	June 17	2 mos.+	clay	1–3-celled; majority 2-celled
April 15...	June 17	2 mos.+	H_2O on filter over cotton	majority 2-celled
April 6...	June 19	2 mos.+	H_2O on filter over cotton	2-celled
May 5...	June 19	1 mo. +	H_2O on filter over cotton	2-celled
May 5...	June 17	1 mo.	soil	2–7-celled (fungus)
May 21...	June 21	1 mo.	sand	2-celled
May 21...	June 21	1 mo.	1% cane sugar filter on cotton	2-celled
May 29...	June 21	1 mo. —	soils	1–4-celled (Chaetomium)
May 29...	June 21	1 mo.	H_2O filter on cotton	2-celled
May 29...	June 21	1 mo.	filter over soil	2–4-celled (fungus)

This rough table shows that the rate of development is variable and slow. The spores of March 23–June 23 were shed in a heap on the moist cotton in the moist chamber containing the plants from the field. Here were found two-celled stages and thalli with branches. Uninfected plants have reached at most 4 and 5 cells, while those with fungi have mature thalli. This difference is apparently due to some change caused by the fungus.

LEITGEB (19) describes the germination of *Aneura pinguis* and *A. palmata*, but figures only the early stages of *A. palmata*. He

says that the spores enlarge strikingly at first, and by one-sided growth a filament is formed which elongates by apical growth, forming a cylindrical body. This body branches and in the tip cell of the main axis and its branches the typical apical cell of the mature thallus arises.

In *Aneura pinguis* the spores at shedding contain chloroplasts as mentioned above (fig. 51). The spore does increase rapidly in size from 60 and 70 μ to 90 and 100 μ in a few days. The plastids are grouped somewhat at one side, where the cell begins to elongate into a slight projection. A wall divides the spore into two unequal cells (fig. 56) (this may happen within 1 or 2 weeks); the smaller one grows until it equals the sister cell. The exospore has not been split, but has elongated and surrounds the two cells (figs. 57, 58). The younger cell is now divided unequally by a vertical wall bent slightly toward the long axis of the cell (figs. 59, 60). It soon grows as large as the cell from which it was cut off, and the division could easily be mistaken for an equal one. This division may also be horizontal, resulting in a dorsal and a ventral cell. The apical cell may originate in either one of these two cells, probably the better lighted one (PEIRCE **22**, LAMPA **16** and **17**, GOEBEL **10–12**, BOLLETER **2**, SCHOSTAKOWITSCH **24**). This second or third wall can then be considered the one which marks out the apical cell.

Only one sporeling was found where the exospore had split and a filament of five cells had grown (fig. 71). The next division comes when the last cell cut off equals that from which it was cut, and the new wall again is a vertical one inclined toward the axis of elongation (fig. 61). This mode of development continues up to the four- and five-celled stage. The only difference between this apical cell and that of the mature thallus is the longer time interval between the segmentation and the division of the segments. In this four- and five-celled stage the echinate projections of the exospore are still present, at a greater distance apart and finally disappearing. The mass of cells looks slightly as has been pictured for *Lejeunia serpyllifolia* (CAMPBELL **3**).

This then reduces *Aneura pinguis* to the condition described by GOEBEL (**12**) for *Metzgeria furcata*, where the filamentous stage or *Vorkeim* consists of one or two cells. The branched filaments are

lacking, which must depend upon the conditions of light and moisture under which they are grown.

Another interesting fact connected with the development of the spore is that the fungus plays some part in it when present. Where the spores fell from the capsule and germinated on the cotton, and in another case where the capsule did not open wide but spores in the line of the valves germinated, a fungus was found infecting the plants. These sporelings were all past the two-celled stage (figs. 64–66). The better lighted thalli were forked, possessing mucilage hairs and rhizoids. In whatsoever way the fungus affects the plant, development at least is hastened. Fungi have been noted in many leafy and some thalloid liverworts (NĚMEC **20,** BOLLETER **2,** GARJEANNE **8,** CAVERS **5**), but only in one case does GARJEANNE note a fungus with the spore, and then only as near it.

The infection begins in any cell of the sporeling (figs. 64–66) and extends irregularly along the lower surface. Large knots of hyphae are found in the cells. At first the cells are not killed, fungus, plastids, and nucleus all being present. Gradually the plastids disappear but the nucleus remains longer. In cells adjoining and near to the infected ones, starch of the plastids has been transformed into dextrine.

A majority of the plants of the field are infected irrespective of habitat. One would like to know whether spores are also infected early or whether the laboratory conditions were such as to favor infection. It is hardly probable that any such relation exists between spores and fungus as BRUCHMANN has found for species of *Lycopodium*. It is more likely, as GARJEANNE thinks, a chance condition, and not at all an endophytic fungus of mycorrhiza plants. Thalli from the field usually have the fungus a short distance behind the actively growing region, and sometimes extending along two-thirds of the dorsal surface. Is it possible that this is one of the main causes for the dying back of the thallus?

Rhizoids are commonly filled with strands of the hyphae (fig. 68). Infection of the rhizoids commonly occurs from the thallus, and when chloroplasts are still present. The elaborate pseudo-parenchyma of fungi described by NĚMEC (**20**) at the base of the rhizoids is lacking, but there are knots of hyphae. Rarely, also,

are the rhizoids as deformed by the fungus as by the obstacles in their path of growth.

Inoculations of pure cultures have not been made because of the desire to get as many sporelings as possible to develop mature thalli. Some of the fungi obtained pure were a species of *Fusarium*, *Cephalothecum roseum*, a species of *Alternaria* and of *Gloeosporium*, and an unidentified one which grew with *Pencillium* in an impure culture. GARJEANNE has found that more than one species may be present at the same time in a rhizoid. It will be interesting to know how many of the above can infect the spores.

Summary

1. The gametophyte of *Aneura pinguis* is a simple, slightly differentiated thallus.

2. Archegonia and antheridia are borne on lateral branches of dioecious plants; they develop according to the *Jungermannia* type.

3. The sporophyte of *Aneura pinguis* is highly specialized. One-half of the embryo at its first division forms a haustorial cell; from the other half capsule, seta, and a temporary foot develop. Sterilization of the tissue of the capsule occurs at three periods: (1) the wall and apical cushion are cut out; (2) the elaterophore is defined; (3) sporogenous tissue is differentiated into elaters and spore mother cells.

4. The capsule splits by four early defined valves. The spores are echinate and contain chloroplasts at maturity.

5. The protonemal stage is reduced to one or two cells. The spore coat incloses the very young sporeling.

6. The mature thallus often contains a fungus. Infection takes place in some sporelings as early as the two-celled stage. Rhizoids may be infected from the thallus.

7. No gemmae are found on *Aneura pinguis*. New plants are produced by the dying back of the old thallus.

Acknowledgments are due Professor JOHN M. COULTER and Professor W. J. G. LAND, under whose direction this work was done.

THE UNIVERSITY OF CHICAGO

LITERATURE CITED

1. BEER, R., On the development of spores of *Riccia glauca*. Ann. Botany **20**:275–291. 1906.
2. BOLLETER, E., *Fegatella conica*. Beih. Bot. Centralbl. **18**:327–408. 1905.
3. CAMPBELL, D. H., The structure and development of mosses and ferns. 1905.
4. CAVERS, F., The inter-relationships of the Bryophyta. III. Anacrogynous Jungermanniales. New Phytol. **9**:108–207. 1910.
5. ———, On saprophytism and mycorhiza. New Phytol. **2**:30. 1907.
6. COKER, W. C., Abnormalities in liverworts. Bryol. **12**:104–105. 1907.
7. EVANS, A. W., Vegetative reproduction in *Metzgeria*. Ann. Botany **24**:271–303. 1910.
8. GARJEANNE, F. M. ANTON, Über die Mykorrhiza der Lebermoose. Beih. Bot. Centralbl. **15**:470–482. 1903.
9. ———, Die Verpilzung der Lebermoosrhizoiden. Flora **102**:147–185. 1911.
10. GOEBEL, K., Archegoniaten studien. 6. Über Function und Anlegung der Lebermoos-Elateren. Flora **80**:1–37. 1895.
11. ———, Über die Jugendzustände der Pflanzen. Flora **72**:15–16. 1889.
12. ———, Organographie der Pflanzen. 1898–1901.
13. HOFMEISTER, W., On the germination, development, and fructification of the higher Cryptogamia. Transl. by F. CURRY. 43–46. 1862.
14. JACK, J. B., Hepaticae Europaeae. Bot. Zeit. **35**:83. 1877.
15. KNY, L., Beiträge zur Entwickelungsgeschichte der laubigen Lebermoose. Jahrb. Wiss. Bot. **4**:6497. 1865–1866.
16. LAMPA, E., Untersuchungen an einigen Lebermoosen. Sitzber. K. Akad. Wiss. Wien **111**:477–487. 1902.
17. ———, Keimung einiger Lebermoosen. Sitzber. K. Akad. Wiss. Wien **112**:779–792. 1903.
18. LE CLERC DU SABLON, Recherches sur le développement du sporogone des Hépatiques. Ann. Sci. Nat. Bot. VII. **11**:126–180. 1885.
19. LEITGEB, H., Untersuchungen über die Lebermoose. 1874–1882. Vol. III. Die frondosen Jungermannieen.
20. NĚMEC, B., Die Mykorrhiza einiger Lebermoose. Ber. Deutsch. Bot. Gesells. **17**:311. 1899.
21. ———, Die Wachstumsrichtungen einiger Lebermoose. Flora **96**:409–450. 1906.
22. PEIRCE, G. J., Studies of irritability in plants: the formative influence of light. Ann. Botany **20**:449–465. 1906.
23. SCHIFFNER, V., Hepaticae in ENGLER and PRANTL's Die natürlichen Pflanzenfamilien **1**:1–144. 1893–1895.
24. SCHOSTAKOWITSCH W., Über die Reproductions- und Regenerations-Eracheinungen bei den Lebermoosen. Flora **79**:350–384. 1894.

EXPLANATION OF PLATES IX–XII

FIG. 1.—Sketch of dioecious thallus.

FIG. 2.—Vertical longitudinal section through apical cell; ×505.

FIGS. 3–8. Serial horizontal section through apical cell; ×830.

FIG. 9.—Horizontal section of thallus, showing two apical cells; ×505.

FIGS. 10, 11.—Horizontal section of thallus showing two apical cells in one sinus; ×830.

FIG. 12.—Young mucilage hairs; ×830.

FIG. 13.—Young rhizoid with chloroplasts present; ×830.

FIG. 14.—Young rhizoid infected by fungus from within; ×830.

FIG. 15.—Rhizoids with and without fungus, showing irregular form; ×175.

FIG. 16.—Vertical section through antheridium initial; ×505.

FIG. 17.—Vertical section, showing antheridium initial divided into stalk and antheridium proper; ×830.

FIG. 18.—Vertical section through antheridium, showing first vertical wall; ×505.

FIG. 19.—Stages of development in antheridium, showing wall and spermatogenous cells defined; ×505.

FIG. 20.—Vertical section through antheridium, showing early divisions of the spermatogenous cells; ×830.

Figs. 21–24.—Horizontal sections through the antheridium, showing its development; ×1650.

FIGS. 25, 26.—Vertical sections through older antheridia, showing stalk cells, and fig. 26 showing the development of the tissue around the antheridium; ×505.

FIG. 27.—Stages of development in the sperm; ×2800.

FIG. 28.—Horizontal section through an archegonial branch, showing numerous archegonia; ×505.

FIG. 29.—Vertical section through young archegonium; ×830.

FIG. 30.—Vertical section through older archegonium; ×830.

FIG. 31.—Horizontal section through archegonial neck; ×830.

FIG. 32.—Vertical section through mature archegonium; ×830.

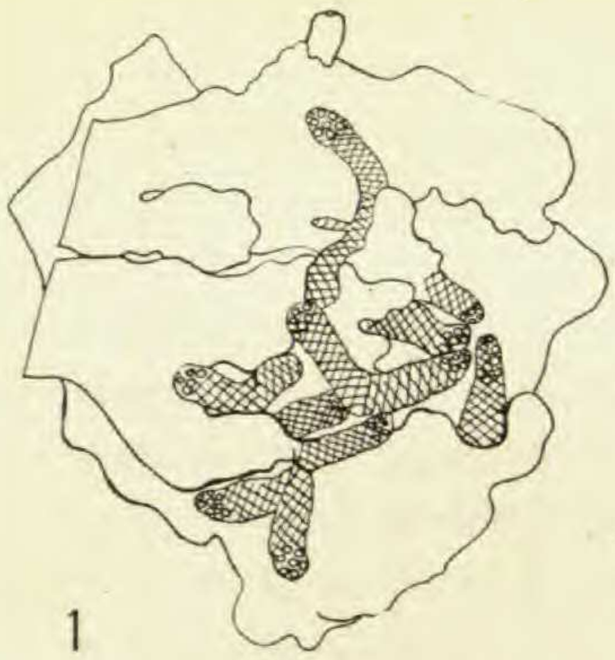
FIG. 33.—Vertical section through archegonium, showing canal cells disorganized; ×830.

FIG. 34.—First division of the young sporophyte; ×1040.

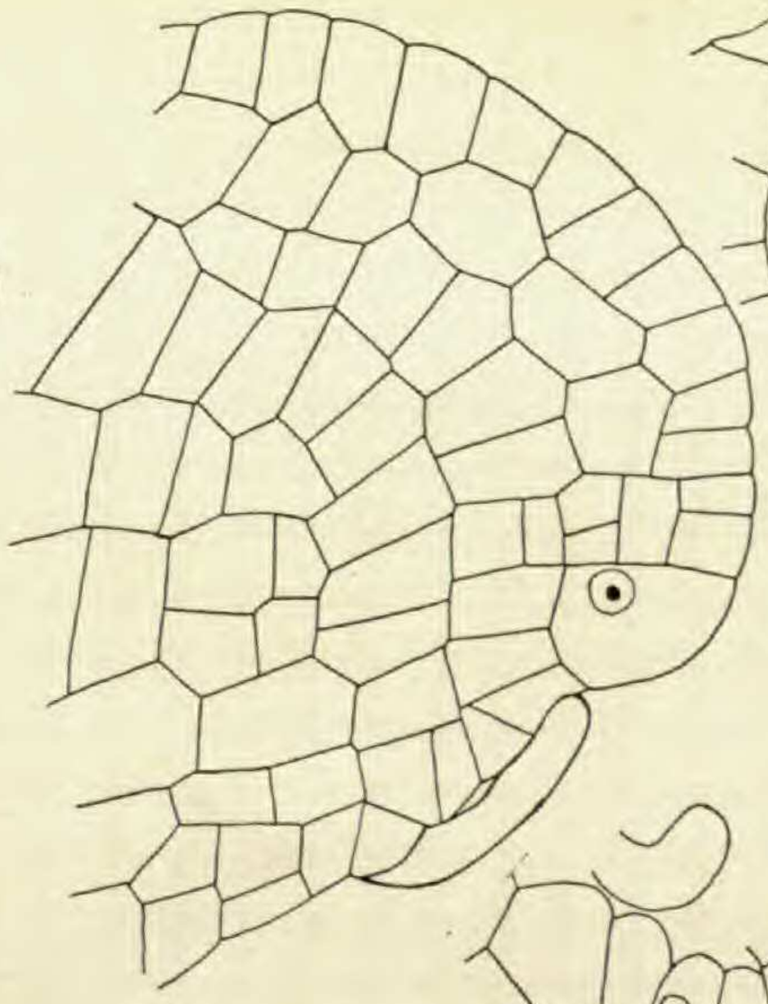
FIG. 35.—The haustorial cell of the sporophyte, well elongated, and gametophyte cells disorganizing; ×1040.

FIG. 36.—Haustorial cell more elongated; the sporophyte proper composed of four cells; ×1040.

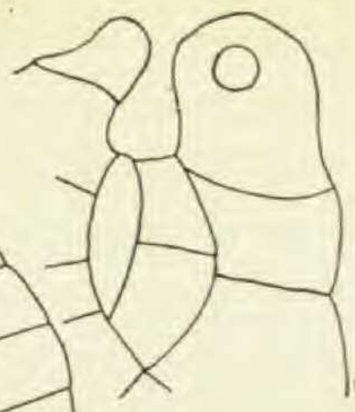
FIG. 37.—Foot, seta, and capsule region of the sporophyte marked out; ×1040.



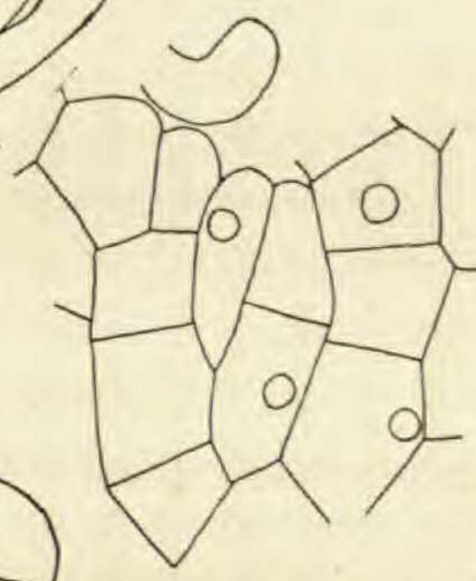
1



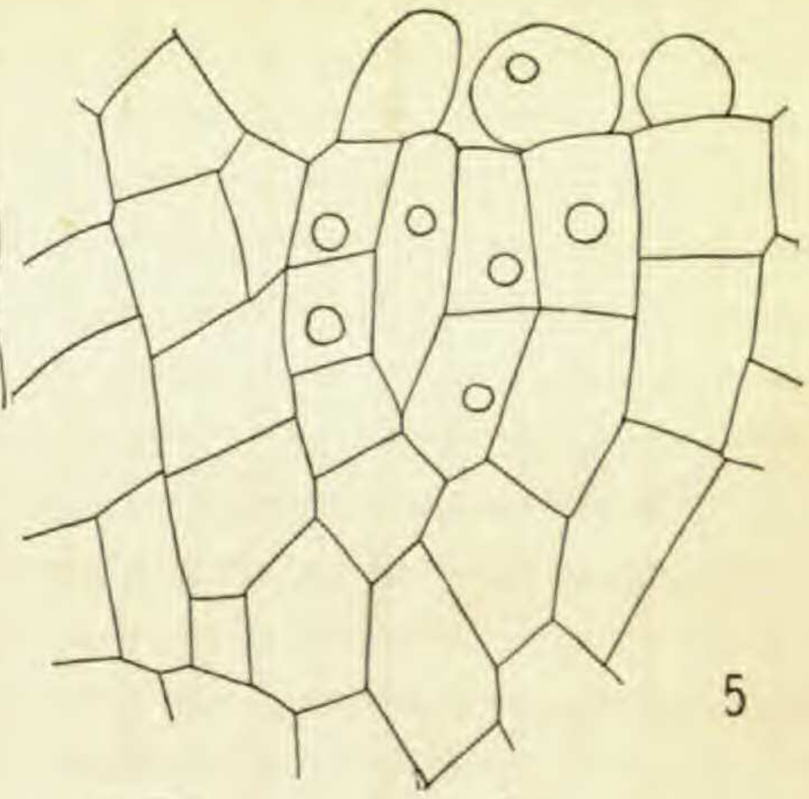
2



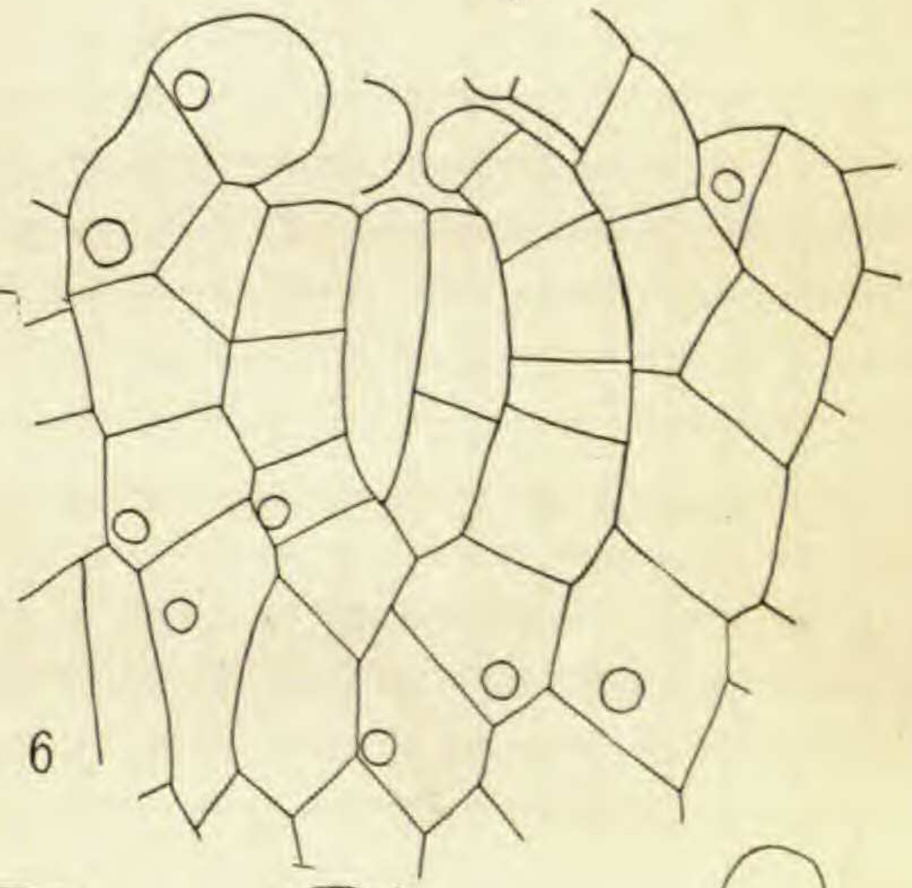
3



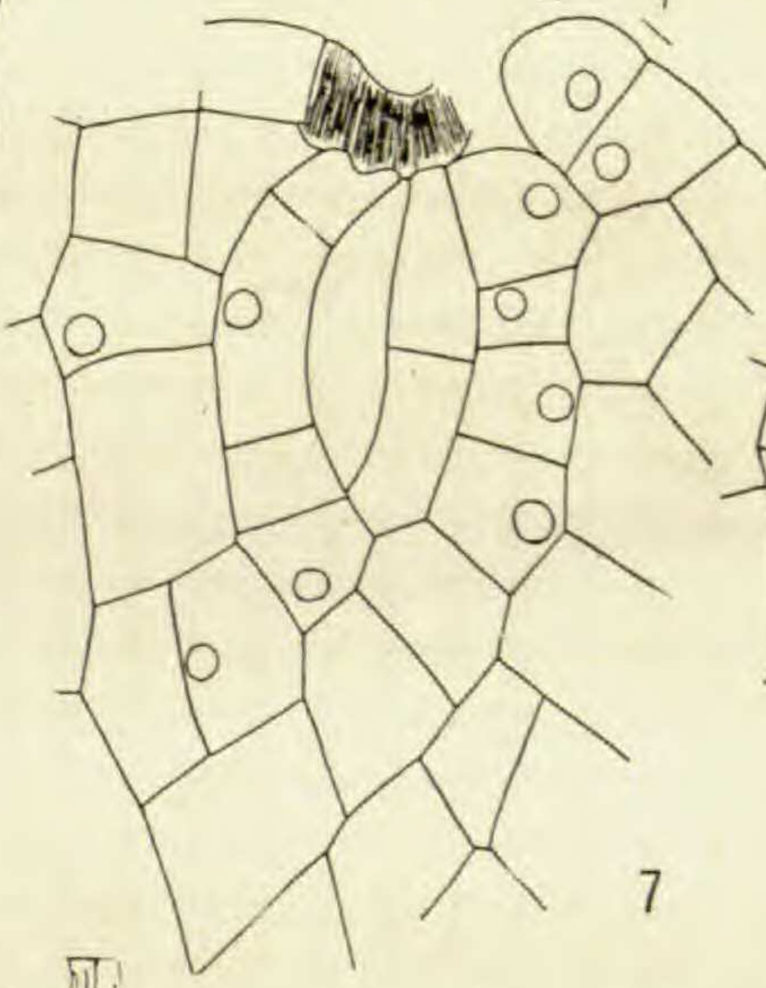
4



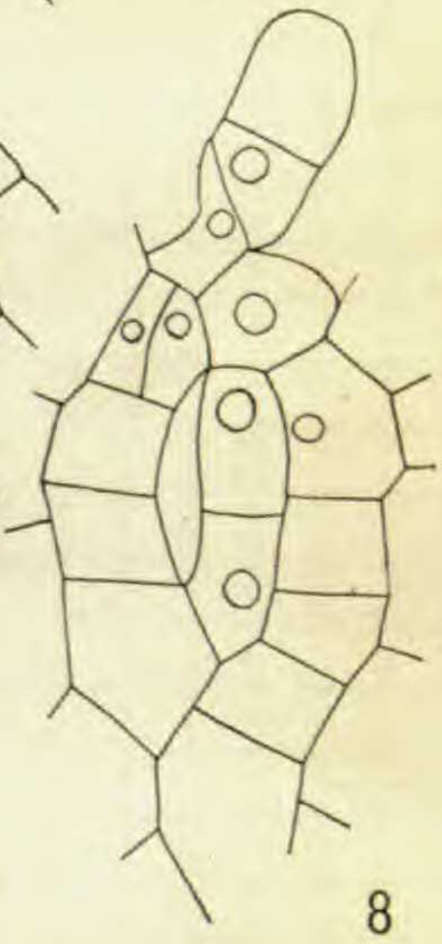
5



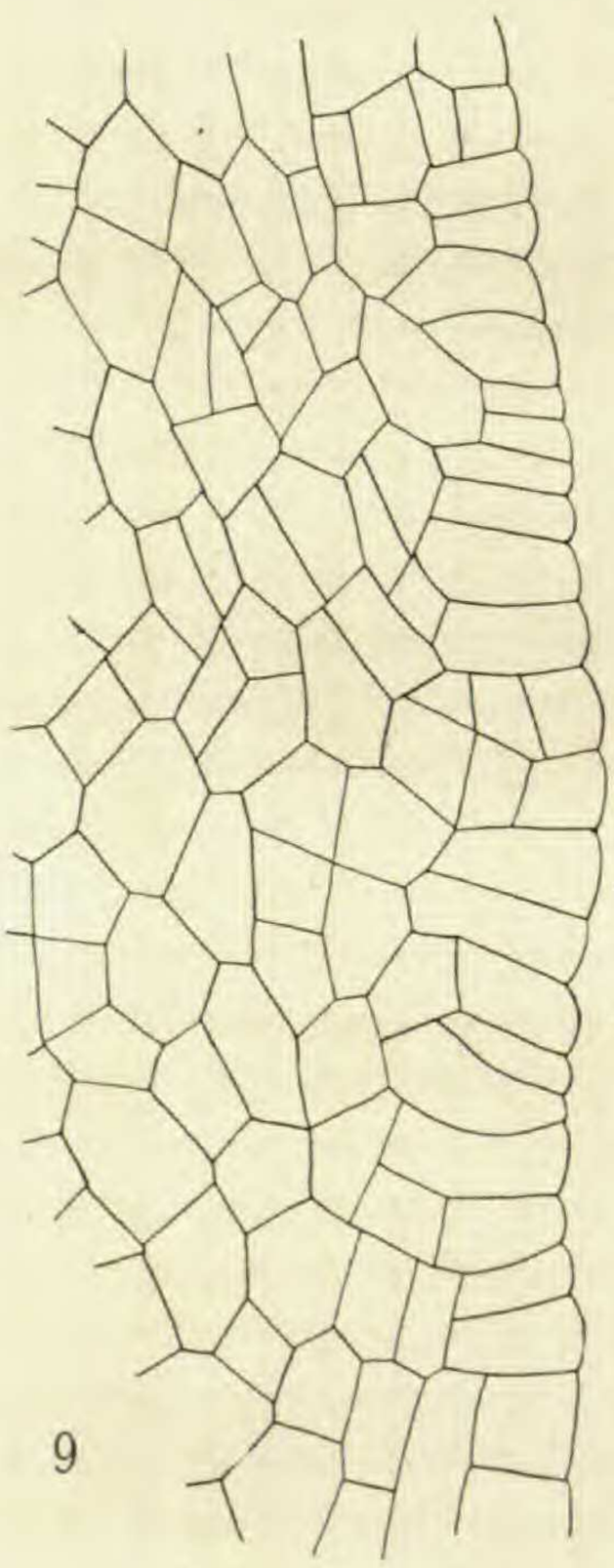
6



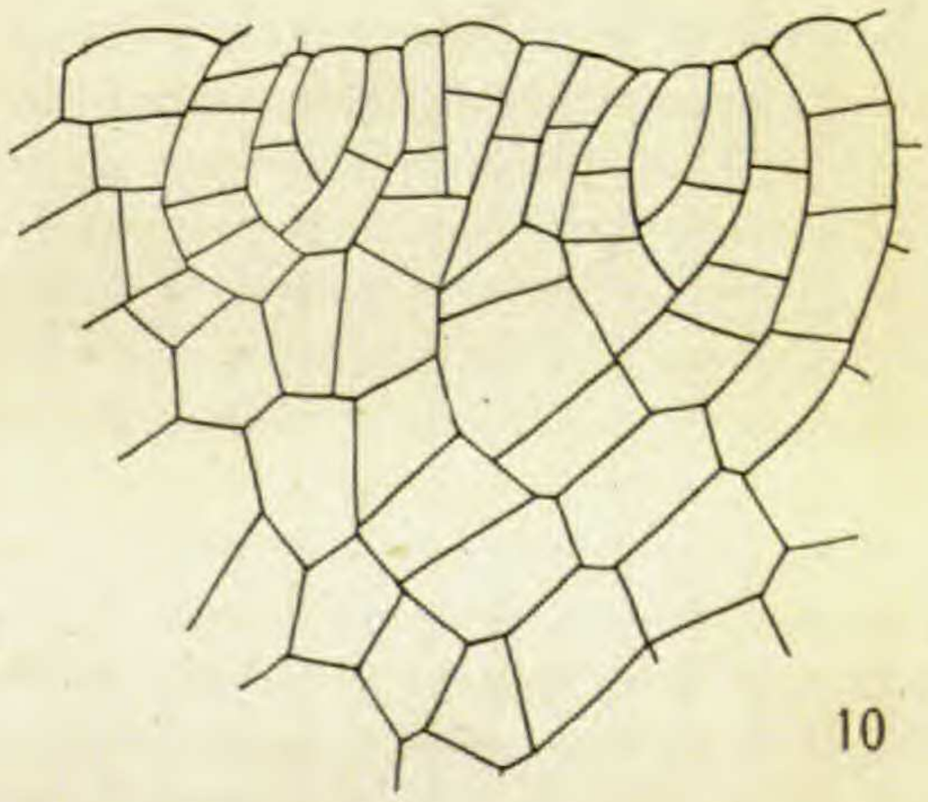
7



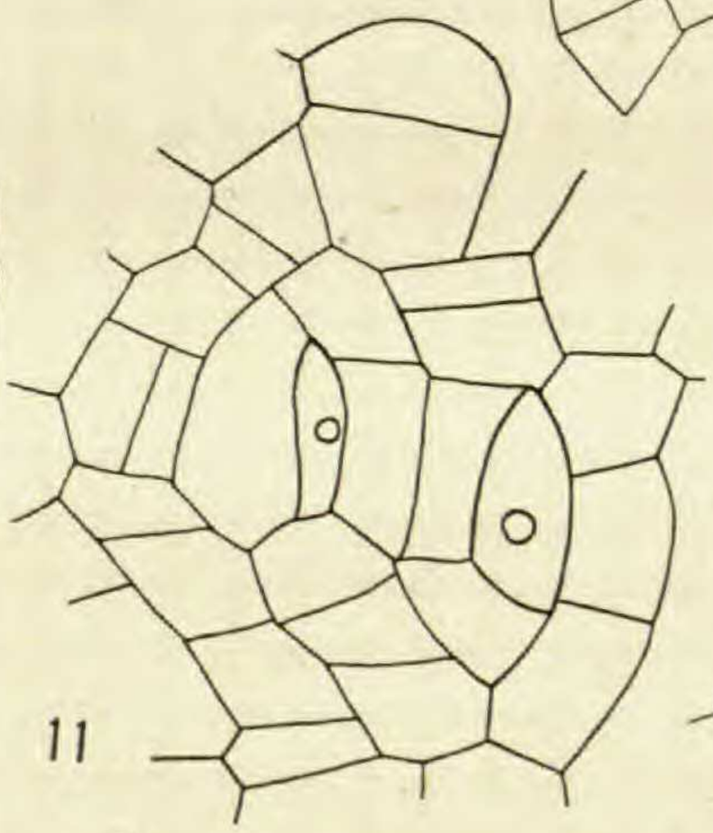
8



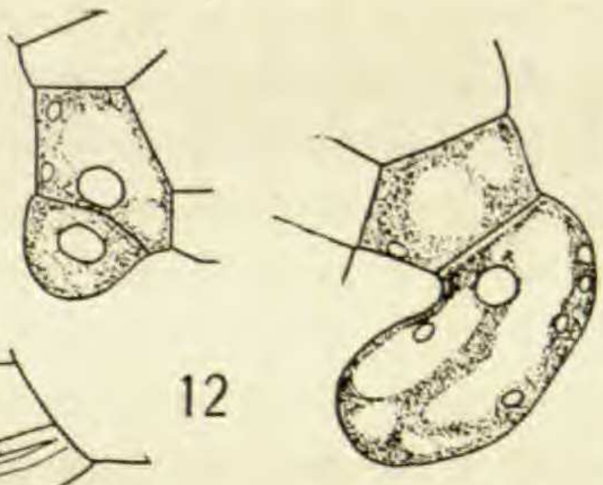
9



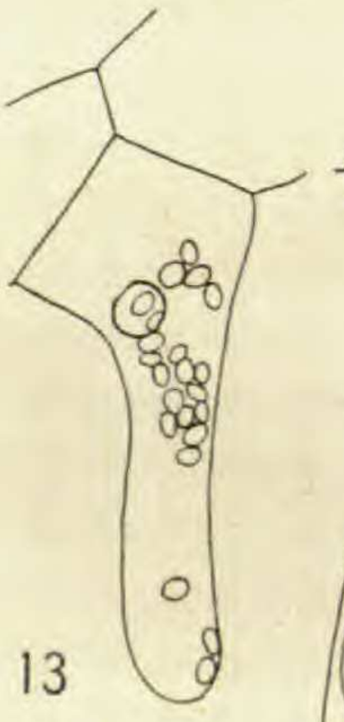
10



11

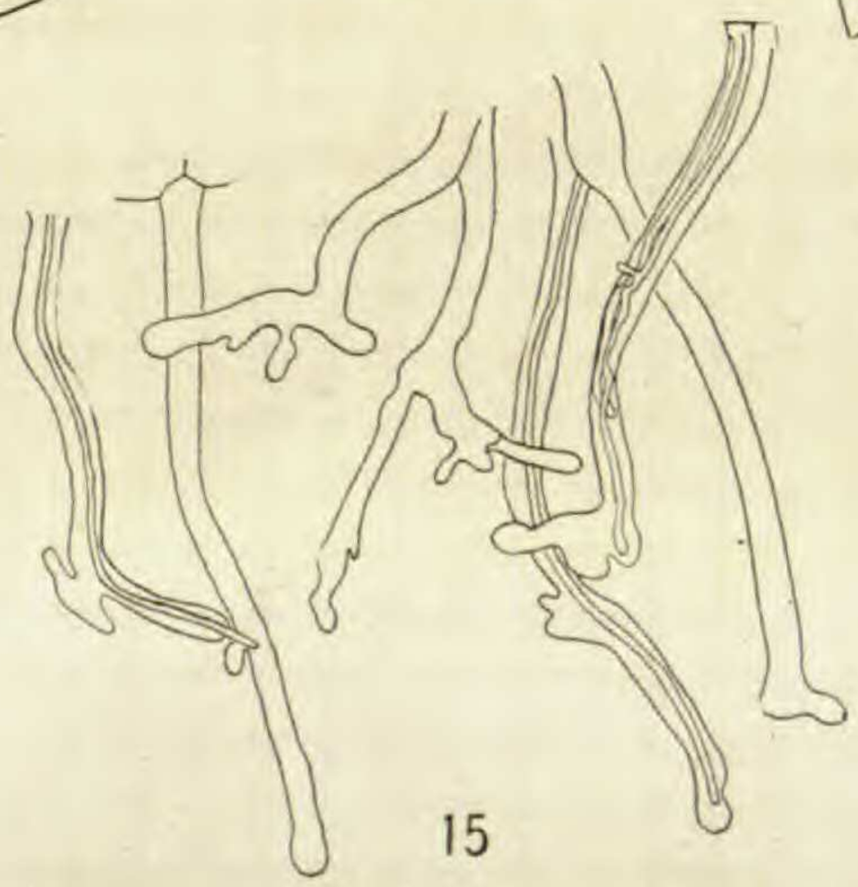


12



13

14



15

G. L. Clapp del.

CLAPP on ANEURA